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VULNERABILITY IN HOLSTEIN-FRIESIAN DAIRY COWS:  
RISK FACTORS FOR CULLING AND EFFECT OF  
TEMPERAMENT ON OESTRUS

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A thesis submitted for the degree of Master of Science by Research



The University of Edinburgh

FEBRUARY, 2011

## **Declaration**

This is to declare that the work presented in this thesis is that of my own and I have not submitted previously to The University of Edinburgh or any establishment for a degree. All other sources of information have been acknowledged by means of references.

Signed : \_\_\_\_\_  
Mr. D. Chiumia

Date : \_\_\_\_\_

## **Dedication**

I dedicate the work to my parents Rev. & Mrs. Chiumia, Kaunda family and all relatives for every support given to me to reach this far through God's grace. "GOD THE EBENEZER"

## **Acknowledgement**

First and foremost, acknowledgement should go to my supervisors: Dr. M.G.G. Chagunda, Dr. D. Roberts and Dr. A.I. Macrae for their unmatched support offered to me during the research work. Never to be taken for granted.

I am grateful to Ross McGinn, Database Manager, Dr. Jenny Gibbons for her advice on approach test technique, Ainsley Bagnall, Paul Kelly, David Bell, staff and technicians at Scottish Agricultural College (SAC) Dairy Research Centre. Bunda College of Agricultural (Animal Science Department) and friends are appreciated for their support rendered in different ways.

Gratefully, I would like to acknowledge funding from The Scottish Government (International Development Fund).

May God shower His blessings on you all!

## Abstract

*The general aims of the work were to determine the factors associated with vulnerability in Holstein-Friesian dairy cows in two scenarios: A) culling and B) the effect of temperament on days to first recorded oestrus in dairy cows. Vulnerability was defined as either an increased risk of being culled or having long interval from calving to first recorded oestrus in cows. The work was carried out in two distinct studies. The objective of the first study was to identify the predisposing factors for an increased risk of culling in adult Holstein-Friesian dairy cows. This study was conducted using data sourced from Scottish Agricultural College Langhill database. Between September 2003 and August 2010, 519 cows calved for the first time and 175 of these were culled. The major reasons for culling were fertility (9.2%), udder problems (9.1%) and accident (6.2%) on which further analysis was performed. The culled cows were matched with their cohorts that survived to a later lactation. Cows assessed for the risk of being culled due to major reasons had a mean age at first calving of 26.2 months (Standard Deviation (SD) =2.4). Cows with high body condition score (BCS) at service and low 60-day (60d) milk protein had a significantly ( $P<0.05$ ) increased likelihood of being culled due to infertility. The regression estimate (RE) was 1.67 for Service BCS and -2.43 for 60d milk protein with predicted probability (PP) of 0.91. However in first lactation heifers, only BCS at service was significant ( $P<0.05$ , RE=2.65 and PP=0.86). Cows with a reduced interval to reach peak milk yield had a significant ( $P<0.01$ ) likelihood of being culled due to udder problems (RE=-0.05 and PP=0.89). Locomotion score and parity were not significant on increasing the risk of culling cows due to accident. Hence higher BCS at service, low 60d milk protein and short duration to peak lactation are factors that place dairy cows at an increased risk of being culled. The second study aimed at determining the association between temperament and days to first recorded oestrus in adult dairy cows. Temperament traits that were studied were flight response score, nervous, interest, shy, bold, fear and docile. Number of days to first recorded oestrus after calving was the dependent variable in the analysis. Seventy Holstein-Friesian dairy cows, mean age at first calving 25 months (SD=1.9) were used in the study. Temperament traits for individual cows were recorded on day 30 before calving, and on days 30 and 60 after calving. Temperament traits were quantified using an Approach Passageway test. On average the first oestrus occurred 55.5 days (SD=17.9) after calving. The study showed that temperament traits did not significantly affect the number of days to first recorded oestrus. Within the temperament traits, nervousness, shyness, boldness, fearfulness and docility were significantly ( $P<0.001$ ) related to flight response score while interest was not. In conclusion, higher than average BCS at service, low milk protein content at day 60 in lactation and short duration to peak lactation exposes cows to an increased risk of being culled while temperament did not influence number of days to first recorded oestrus after calving.*

## Table of Contents

<b>DECLARATION.....</b>	<b>II</b>
<b>DEDICATION.....</b>	<b>III</b>
<b>ACKNOWLEDGEMENT.....</b>	<b>IV</b>
<b>ABSTRACT .....</b>	<b>V</b>
<b>TABLE OF CONTENTS.....</b>	<b>VI</b>
<b>LIST OF TABLES .....</b>	<b>IX</b>
<b>LIST OF FIGURES .....</b>	<b>X</b>
<b>CHAPTER 1 .....</b>	<b>1</b>
<b>GENERAL INTRODUCTION.....</b>	<b>1</b>
<b>1.1 INTRODUCTION.....</b>	<b>2</b>
<b>1.2 STUDY JUSTIFICATION.....</b>	<b>3</b>
1.2.1 AIMS OF THE STUDY.....	4
<b>1.3 LITERATURE REVIEW.....</b>	<b>4</b>
<b>CHAPTER 2 .....</b>	<b>9</b>
<b>CULLING IN HOLSTEIN-FRIESIAN DAIRY COWS.....</b>	<b>9</b>
<b>2.1 INTRODUCTION.....</b>	<b>10</b>
2.1.1 GENERAL OBJECTIVE .....	10
2.1.1.1 <i>Specific Objectives</i> .....	11
<b>2.2 MATERIALS AND METHODS .....</b>	<b>11</b>
2.2.1 ANIMALS.....	11
2.2.2 DATA.....	12
2.2.2.1 <i>Selection of traits</i> .....	13
2.2.2.1.1 Traits evaluated for cows culled due to infertility .....	13
2.2.2.1.2 Traits evaluated for cows culled due to udder problems .....	15
2.2.2.1.3 Traits evaluated for cows culled due to accident .....	15
2.2.3 STATISTICAL ANALYSIS.....	16

<b>2.3</b>	<b>RESULTS .....</b>	<b>17</b>
2.3.1	DESCRIPTIVE STATISTICS .....	17
2.3.2	REASONS FOR CULLING .....	17
2.3.3	PREDISPOSING FACTORS FOR COWS CULLED FOR EACH OF THE THREE MAIN REASONS .....	20
2.3.3.1	<i>Cows culled due to infertility</i> .....	20
2.3.3.2	<i>Cows culled due to udder problems</i> .....	23
2.3.3.3	<i>Cows culled due to accident</i> .....	25
<b>2.4</b>	<b>DISCUSSION .....</b>	<b>26</b>
<b>2.5</b>	<b>CONCLUSION.....</b>	<b>33</b>
<b>CHAPTER 3 .....</b>	<b>34</b>	
<b>EFFECT OF TEMPERAMENT ON DAYS TO FIRST RECORDED</b>		
<b>OESTRUS IN DAIRY COWS .....</b>		
<b>3.1</b>	<b>INTRODUCTION.....</b>	<b>35</b>
3.1.1	GENERAL OBJECTIVE .....	36
3.1.1.1	<i>Specific Objectives</i> .....	36
<b>3.2</b>	<b>MATERIALS AND METHODS .....</b>	<b>36</b>
3.2.1	ANIMALS .....	36
3.2.2	EXPERIMENTAL DESIGN AND DATA .....	37
3.2.3	STATISTICAL ANALYSIS .....	38
3.2.3.1	<i>Temperament Traits</i> .....	38
3.2.3.2	<i>Temperament and oestrus</i> .....	39
<b>3.3</b>	<b>RESULTS .....</b>	<b>39</b>
3.3.1	DESCRIPTIVE STATISTICS .....	39
3.3.1.1	<i>Flight Response Score</i> .....	40
3.3.2	TEMPERAMENT TRAITS .....	43
3.3.3	TEMPERAMENT AND OESTRUS .....	44
<b>3.4</b>	<b>DISCUSSION .....</b>	<b>45</b>
<b>3.5</b>	<b>CONCLUSION.....</b>	<b>49</b>
<b>CHAPTER 4 .....</b>	<b>50</b>	
<b>GENERAL DISCUSSION .....</b>		
		<b>50</b>



<b>4.1</b>	<b>VULNERABILITY IN DAIRY COWS .....</b>	<b>51</b>
	<b>REFERENCES.....</b>	<b>53</b>
	<b>APPENDIX .....</b>	<b>60</b>
A.	APPROACH TEST PROTOCOL.....	60

## List of Tables

Table 1-1: International body condition scoring systems .....	6
Table 2-1: Reasons for culling in Holstein-Friesian dairy cows.....	18
Table 2-2: Number of cows culled per lactation in Holstein-Friesian dairy herd.....	19
Table 2-3: Descriptive statistics of cows evaluated for increased risk of culling cows due to infertility.....	20
Table 2-4: Likelihood estimates for the increased risk of culling cows due to infertility.....	21
Table 2-5: Odds Ratio of the traits included in the model used to predict the increased risk of culling cows due to infertility.....	21
Table 2-6: Likelihood estimates for the increased risk of culling first lactation heifers due to infertility.....	22
Table 2-7: Odds Ratio of the traits included in the model used to predict the increased risk of culling first lactation heifers due to infertility.....	22
Table 2-8: Descriptive statistics of some of the traits evaluated for increased risk of culling cows due to udder problems.....	23
Table 2-9: Likelihood estimates for the increased risk of culling cows due to udder problems.....	24
Table 2-10: Odds Ratio of the traits included in the model used to predict the increased risk of culling cows due to udder problems. ....	24
Table 2-11: Descriptive statistics of cows evaluated for increased risk of culling cows due to accident. ....	25
Table 3 -1: Descriptive statistics of some of the traits studied in the Langhill herd..	39
Figure 3-2: Flight response score averages at day -30, +30 and +60 from calving...	41
Table 3-2: Spearman's rank correlation coefficients of temperament traits.....	43
Table 3-3: Probability values of the independent variables in the model used to determine the effect of temperament on oestrus. ....	44
Table A-1: The flight response score used to score the cow's flight response to the AT test.....	61
Table A -2: Qualitative terms and descriptions used in the AT test. ....	62

## List of Figures

Figure 1-1: The acceptable range of Body Condition Score (BCS) profiles that allows optimal production of dairy cows without compromise to their reproduction, health and welfare. ....	7
Figure 3-1: Distribution of animals on 9-points flight response score ordinal scale. ....	40
Figure 3-2: Flight response score averages at day -30, +30 and +60 from calving... ..	41
Figure 3-3: Comparisons of flight response scores between primiparous and multiparous cows at time A (1), B (2) and C (3). ....	42

## **Chapter 1**

### **General Introduction**

## **1.1 Introduction**

This thesis presents research work describing factors associated with vulnerability in Holstein-Friesian dairy cows. Vulnerability in the current work was defined as either an increased risk of being culled or having a long interval from calving to first recorded oestrus in dairy cows. Studies on culling indicate that reproductive problems are among the major reasons for culling cows in dairy herds before finishing their productive life (Bascom and Young, 1998; Cozler et al., 2009; Esslemont and Kossaibati, 1997 ; Milian-Suazo et al., 1988). The reproductive failure in high yielding breeds has been associated with factors such as oocyte and embryo quality (Leroy et al., 2008) and high body condition loss (Knop and Cernescu, 2009). Wathes et al., (2007) stated that high body condition loss which is one of the indicators of cows being in negative energy balance affects the hormonal system resulting in low conception rates. Besides energy deficit, Lopez et al., (2007), indicated that the risk of culling first parity cows in later lactations is increased if the cows had a more difficulty calving. Difficult calving in dairy cows has a negative impact on cow performance because it impairs reproductive performance (Lopez et al., 2007; Price and Wiltbank, 1978). Other challenges affecting fertility levels in dairy cows include metabolic disorders (for instance ketosis) (Gillund et al., 2001) and stress (Waiblinger et al., 2004). Cows with a history of ketosis before service have been reported to have a decreased likelihood of conception to that service (Gillund et al., 2001). Rough handling of cows at service which results in increased stress has been associated with lowered conception rate. Some authors (Hilary and Smith, 2000) also argue that there is evidence of social stressors affecting fertility in dairy cows. The latter suggested that cows with a high social status in dairy herd tended to be more fertile.

Fertility affects culling in dairy cows through its influence on days open (the interval between calving and successful service) and calving interval (interval between successful calvings) (Bousquet et al., 2004). Poor fertility further results in lower average annual milk yield per cow, high replacement and culling rates (Cozler et al., 2009; Garnsworthy, 2004). Additionally, dairy cows are also culled because of

diseases, udder problems, accident, production and feet and leg problems (Bascom and Young, 1998; Cozler et al., 2009). For example Milian-Suazo et al., (1989) reported a high risk of culling cows due to udder problems if the cows had higher current milk yield per day, clinical mastitis or teat problems. Rushen and Passille (2006) reported an increased incidences of lameness and hoof problems in dairy cows kept in houses with concrete floors thus showing how housing condition can contribute to culling in dairy herds.

Temperament of cows is another factor which is associated with their reproduction performance (Grandin, 2003; Phocas et al., 2006). Temperament is often described as an individual trait influencing an animal's behavioural response to handling (Gibbons et al., 2009). According to Phocas et al., (2006), genetic estimates between temperament and reproduction traits showed a positive correlation. For instance, there has been an indication of less fearful heifers being more productive as measured by high reproduction and calving performance (Phocas et al., 2006). On a physiological basis, stress related factors for example anticipation of danger stimulates secretion of catecholamines (epinephrine and norepinephrine) as part of the fight-or-flight reaction (Robert and Mathew, 2000). Ines et al., (2002) showed that catecholamines can influence maturation of oocytes because these hormones affect the metabolic behaviour responsible for maturation of oocytes. Thus behavioural characteristics of animals have the potential to be related to the susceptibility of animals to different reproductive challenges.

## **1.2 Study Justification**

Despite the need for genetic improvement of dairy cows, fertility has deteriorated as a negatively correlated response to selection for higher milk yield (Lovendahl and Chagunda, 2006). In addition Gibbons et al., (2009) suggested that there might be possible undesirable consequences on cow temperament that have been inadvertently selected for by the predominant selection for production traits in breeding programmes. Both infertility and the temperament of cows have been associated with various indicators of reduced reproductive performance (Cozler et al., 2009;

Waiblinger et al., 2004; Wathes et al., 2007) which increase involuntary culling rates on dairy farms, hence affecting dairy farming profitability. The reasons for culling have been well documented (Bascom and Young, 1998; Cozler et al., 2009) but factors that increase the vulnerability of dairy cows to culling before finishing their productive life in different dairy systems are sparse. Hence the need to quantify factors that increase vulnerability of dairy cows to an increased risk of being culled and having a delay in onset of first oestrus after calving.

### **1.2.1 Aims of the Study**

The general objective of the work presented in this thesis was to determine factors associated with vulnerability in Holstein-Friesian dairy cows in two scenarios:

1. Culling in Holstein-Friesian dairy cows.
2. The effect of temperament on days to first recorded oestrus in Holstein-Friesian dairy cows.

## **1.3 Literature Review**

The dairy cow is the core unit of the dairy industry. Understanding the culling process in dairy herds and its consequences is therefore extremely important in order to optimise production (Hadley et al., 2006). However, selection for high milk production has resulted in undesirable effects on cow fertility (Lovendahl and Chagunda, 2006). The susceptibility of dairy cows to fertility challenges has been associated with various risk factors such as a decrease in conception rate at first service (Bousquet et al., 2004). Literature suggests that one of the contributing factors is energy deficit during early lactation (Maurice and Lonergan, 2003; Vries and Veerkamp, 2000; Wathes, 2010). Energy deficit comprises an imbalance between diet energy input and production requirements (Ross et al., 2008). Some authors add that the risk for increased susceptibility of cows to enter severe negative energy balance is related to over-conditioning of high producing dairy cows (Rukkwamsuk et al., 1999). There are various changes in biochemical, endocrine system and metabolic pathways which result from negative energy balance, and these

are associated with different signs of reduced fertility; for example, a delay in the first visible signs of a cow being on heat after calving. According to Roche et al., (2009), in response to a period of chronic energy deficit, key hormone expression and tissue responsiveness to hormone levels change. These changes increase lipolysis and decrease lipogenesis processes, in order to provide much need energy for milk production. This is the reason that the transition period from dry period to lactation is a critical time for dairy cows (Minor et al., 1998). From an economic point of view, inefficiency in reproductive performance is associated with a long calving interval which is a consequence, among other factors, from a failure to detect oestrus (Heuwieser et al., 1994). Previous studies (Leroy et al., 2008; Maurice and Lonergan, 2003; Mulligan et al., 2007) suggest a connection between negative energy balance and ovarian function. Dairy cows enter into a state of negative energy balance (Mulligan et al., 2007) when feed intake is unable to support the required energy for milk yield and maintenance (Vries and Veerkamp, 2000). According to Knop et al., (2009), during early lactation cows do not eat as much as compared to the second or third month of lactation. The challenge in dairy cows is that when they experience severe negative energy balance, it results in a reduction in luteinising hormone (LH) pulse frequency and circulating concentration of both insulin and insulin-like growth factor-1 (IGF-1) (Leroy et al., 2008; Mulligan et al., 2007). Hormones such as growth hormone (GH), insulin and IGF-1 all have direct influence on ovarian function (Wathes et al., 2007). In addition, there is a strong correlation between negative energy balance and first postpartum ovulation (Maurice and Lonergan, 2003) hence the delay in onset of first oestrus after calving.

A number of reports have shown additional challenges that may have detrimental effects on both cow reproductive performance and/or production. These challenges include endometritis (Back et al., 2009; Hajurka, 2009; Petrujkic' et al., 2008), lameness (Green et al., 2002; Roger and Seifollah, 2007), mastitis (Piepers et al., 2009; Wathes, 2010), retained placenta (Ganah et al., 2008), ketosis (Duffield, 2010) and ovarian cysts (Vacek et al., 2007). For example, endometritis leads to prolongation of service period and a high insemination index of affected cows (Petrujkic' et al., 2008). Severe lameness can reduce milk yield both before and after



it is diagnosed and treated (Green et al., 2002). On the other hand, according to Piepers et al., (2009), heifers calving with either subclinical or clinical mastitis tend to be culled in their first lactation. Evidence also shows that incidence of mastitis is increased as a function of machine milking. For instance, Chagunda et al., (2006) used duration of milking as an index reflecting the negative effect of machine milking on teat integrity. In the latter study, the assumption used was that the longer the duration of milking, the higher the risk of mastitis incidence. This agrees with Hovinen and Pyörälä (2010) who reported a general deterioration in udder health in dairy cows following the introduction of automatic milking.

However, managerial activities can be put in place to mitigate the impact of some of the risk factors outlined. One such routine managerial activity is body condition scoring which is a method of assessing animal body energy reserves (Edmonson et al., 1989). Due to differences in methods of body condition scoring animals, various systems have been developed (see Table 1-1). This system of estimating fat reserves is used as a tool to aid feeding management (Ferguson et al., 1994). To achieve good feeding efficiency, adjustments can be made in respect to herd nutrition status (Waltner et al., 1993).

Table 1-1: International body condition scoring systems

Country	Scale	Interval (points)	Visual or palpation
United Kingdom, Ireland	0 to 5	0.5 (11)	Palpation
United States	1 to 5	0.25 (17)	Visual
New Zealand	1 to 10	0.5 (19)	Palpation
Australia	1 to 8	0.5 (15)	Visual
Denmark	1 to 9	1 (9)	Visual

Source: Bewley and Schutz, (2008)

Apart from improving feeding efficiency, body condition scoring helps to ensure that cows achieve optimal body condition which is required during each stage of the lactation period in order to have optimal productivity (Wildman et al., 1982).

Through body condition scoring, farmers make sure that cows are not over-conditioned especially at calving (Waltner et al., 1993). Lower body condition score (BCS) is associated with reduced reproduction and production, whilst excessive body condition score ( $BCS \geq 3.5$ , 5-point scale) is associated with a reduction in dry matter intake and an increased risk of metabolic disorders (Roche et al., 2009). In addition, Roche et al., (2009) suggested a range of BCS profiles (shown in Figure 1-1) that can allow dairy cows to optimise milk production without detrimental effects on their reproduction, health and welfare.

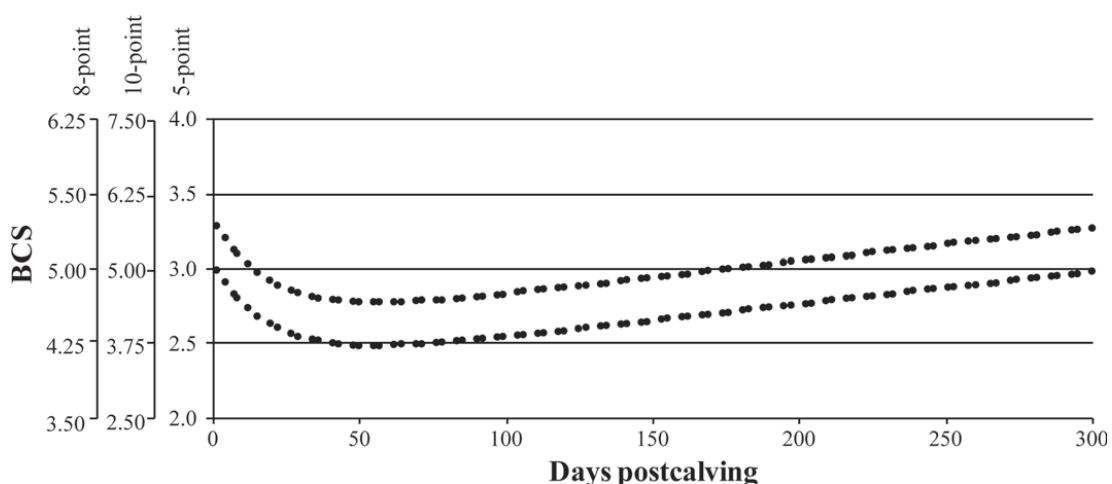


Figure 1-1: The acceptable range of Body Condition Score (BCS) profiles that allows optimal production of dairy cows without compromise to their reproduction, health and welfare.

Source: Roche et al., (2009).

The Figure 1-1 shows that after calving, there is general body condition loss in dairy cows but gradually their condition improves later in the lactation as indicated by the top dotted line (maximum BCS) and the bottom dotted line (minimum BCS). However to reduce the negative feedback effect of BCS loss on subsequent fertility, Garnsworthy et al., (2008) suggested minimising the loss of BCS at calving, and indicated that high genetic merit cows are capable of maintaining BCS of 2.5 (5-point scale) throughout lactation. One of the arguments proposed is that the mean biological target BCS at calving has decreased with genetic improvement for milk production, such that modern dairy cows are genetically thinner.

Cow comfort and cow housing are some of the important aspects when looking at issues to do with animal welfare (Rushen and de Passille, 2006). Concrete floors for instance may impair locomotion, increase the risk of injury and influence expression of oestrus behaviour (Rushen and de Passille, 2006). In addition Rousing et al., (2005) reported that there is decreased welfare in cows which are more susceptible to increased morbidity and mortality, and decreased production. The need to improve the quality of animal-human relationship is therefore important. Positive and gentle interaction of humans and animals is vital as it reduces the risk of accidents to the handler (Rousing et al., 2005; Waiblinger et al., 2004). Contrary, if cows are not showing defensive reactions or startling at handling, there is an reduced risk of injuries to the animals themselves (Waiblinger et al., 2004). Rough handling might result in reduced performance, and in general both physiological and behavioural stress reactions affect cow performance (Waiblinger et al., 2004). According to Rousing et al., (2005), fear of humans can be a cumulative source of stress in animals because animals showing fearful behaviour often inappropriately react to handling situations. The negative side of rough handling at service is that it increases plasma adrenalin and lowers conception rate (Waiblinger et al., 2004). The added importance of improving the human-animal relationship is that, while attempts are under way to improve the welfare of animals, the safety of the humans involved in handling must also be considered. To achieve the desired human-animal relationship in dairy cows, human avoidance and approach tests have been proposed as means of assessing the quality of the relationship (Rousing et al., 2005), with a view to optimising dairy production (Waiblinger et al., 2004).

However effects of selective breeding seem to have affected temperament in dairy cows (Gibbons et al., 2009), which affects the ease of handling for example during veterinary procedures (Waiblinger et al., 2004). It was suggested by Gibbons et al., (2009) that while it is important to improve production traits, it is also valuable to consider the effects of breeding programmes on cow temperament. Hence it has been recommended by Gibbons et al. (2009) to use the approach test as one of the methods to measure specific temperament traits in dairy cows.

## **Chapter 2**

### **Culling in Holstein-Friesian Dairy Cows**

## **2.1 Introduction**

Most of the health costs incurred on a dairy farm are associated with increased labour required at delivery, increased number of days open, death of cows and calves, and culling of cows (McGurk et al., 2007). Thus culling practices should be scrutinised to optimise milk production profits, as well as a prerequisite for better understanding of the culling process and its consequences (Hadley et al., 2006). Culling is the act of identifying and removing a cow from a herd, and assuming a constant or expanding herd size, replacing the cow with another cow, often a first lactation heifer (Hadley et al., 2006). In farm animals this means disposal, because culling increases the need for more replacements to maintain the herd size. Generally two classes of culls exist: involuntary culls and voluntary culls. A previous study by Whitaker et al., (2004) indicated that half of the cows culled during a four year period were culled for involuntary reasons. Some of the reasons for culling dairy cows as reviewed by other studies include; reproduction, poor production, health, age, calving difficulties, death, accident and replacement (Bascom and Young, 1998; Cozler et al., 2009). However it is important to recognise that culling is done for many reasons, including the individual farm or farmer's goals. From a physiological basis, low IGF-1 concentrations in postpartum dairy cows have been associated with increasing culling rates due mainly to poor fertility (Lyons et al., 2009). In some cases potential heifers are lost through perinatal mortality, especially in situations where calving assistance was required (Brickell et al., 2009). In addition, death of young stock or failure to conceive prevent replacement heifers from even reaching their first lactation, and that approximately one-third of all animals that do calve complete only a single lactation (Wathes et al., 2007). However, studies examining the predisposing factors and characteristics of dairy cows that do not progress to the end of their productive life are sparse.

### **2.1.1 General Objective**

The overall aim of this study was to identify the predisposing factors for increased risk of culling in adult Holstein-Friesian dairy cows.

### **2.1.1.1 Specific Objectives**

The specific objectives were:

1. To identify the main reasons of culling in Holstein-Friesian dairy cows in one UK dairy herd.
2. To identify the predisposing factors for increased risk of culling due to the three main reasons for culling in Holstein-Friesian dairy cows.

## **2.2 Materials and Methods**

### **2.2.1 Animals**

Data were obtained from the Scottish Agricultural College (SAC) database. The data were of the Langhill herd of Holstein Friesian cows, which were on a long-term genetic and feeding systems project as described by Bell and Roberts, (2006). Cows were either of the genetic line selected for kilograms of fat plus protein (select line) or selected to remain close to the average genetic merit for fat plus protein production for all animals evaluated in the UK (control line) (Bell and Roberts, 2006). Both lines were fed either a high-or low-forage diet as a total mixed ration (TMR) (Chagunda et al., 2009). In the high forage management system, cows were fed a complete diet containing between 70% and 75% forage on a dry matter basis, while the diet for low forage cows contained between 45% and 50% forage on a dry matter basis (Chagunda et al., 2009). The TMR were formulated to contain approximately 1200 kg concentrate per lactation for the low forage system, and 3000 kg concentrate per lactation for the high forage system. The long-term genetic and feeding systems therefore had four groups (production systems) based on genetic merit and feeding system of the cows. The production systems were: high forage control (HFC), high forage select (HFS), low forage control (LFC) and low forage select (LFS). In addition, all cows on the high forage diet were at grass during the summer, while those on the low forage diet were housed throughout the year. Cows were milked three times a day and individual body weights were recorded after every milking time. The body condition of the cows was assessed by a five-point scoring

system described by Ferguson et al., (1994) on a weekly basis. Emaciated cows were scored 1; thin cows, 2; average cows, 3; fat cows, 4; and obese cows, 5. Locomotion scoring was done using a 5-point system detailed by Sprecher et al., (1997) to detect lameness in the herd. Body condition and locomotion scoring were carried out at the same time. The clinical description for each locomotion scoring scale point was; normal, mildly lame, moderately lame, lame and severely lame for 1,2,3,4 and 5, respectively. Samples of milk were collected at fortnightly intervals for somatic cell count (SCC), milk fat and milk protein content analysis. Any cow removed from the herd either voluntarily or involuntarily was recorded as culled with the reason recorded. Where the actual reason(s) for culling a cow could not be established, the terms “Unknown” or “Others” were used.

### **2.2.2 Data**

Data used for the current analysis were recorded from September 2003 to August 2010. Cows studied were those which had their first calving within the study period. Cows culled during the period were classified depending on the reason(s) recorded in order to identify the main reasons for culling in the sampled Holstein-Friesian herd. Animals culled because of not seen in heat, were repeat breeders and had reproductive disorder were classified as being culled for infertility. Udder problems comprised the following reasons for culling: lost quarters, mastitis, high SCC, damaged udder and poor udder conformation. Cows culled because they got injured accidentally were part of the group classified as accident. Further information on this group of cows was sourced through oral communication. Lameness and any injury associated with the foot were classified as foot or leg problems. Undefined reasons were included in a class called Unknown or Others. After arranging the classified reasons in a descending order of the total number of cows culled per reason, major reasons for culling were identified.

Having identified the major reasons for culling in the herd, further analysis to identify factors that predisposed cows to an increased risk of culling due to the three major reasons identified was conducted. In this analysis, the culled cows were

matched with their cohorts that survived in the same lactation in which the cow was culled. The control matching was based on age at first calving, feeding system, genetic line and lactation number. Each pair comprised cows which were under the same production system, had the same lactation number and equal or closest possible age at first calving. Ideally the cows were under the same production system for three lactations, but there were very few cows with at least three lactations in the herd. Blocking age at first calving, feeding system, genetic line and lactation number aimed at controlling their influence on the fate of culling a cow, so that the effect of culling would be highly influenced by the traits which were evaluated. The analysis was conducted separately for each of the identified major reasons for culling. Further analysis was done on first lactation heifers culled due to infertility because fertility was identified as the predominant reason for culling first lactation heifers

#### **2.2.2.1 Selection of traits**

Traits associated with culling for infertility, udder problems and accident in dairy cows were selected for further evaluation. Initially all the traits were assessed for possible correlation before inclusion in the statistical model. The prior assessment aimed at preventing multicollinearity which causes the signs on the regression coefficients to be opposite from that which is expected (Moskowitz and Wright, 1985). For instance, if one is associating a decreased secretion of adrenaline hormone with factors such as anticipation for danger, it would be expected that these traits would be inversely correlated. Such prior assessment resulted in only one of any two highly correlated traits being used in the subsequent analysis. Following is the description of the traits included in the final models fitted for each of the three main reasons for culling.

##### **2.2.2.1.1 Traits evaluated for cows culled due to infertility**

Eight traits were evaluated for an increased risk of culling cows due to infertility. The traits were: age at first calving, metabolic calving weight, calving ease, BCS at service, genetic line, feeding system, mastitis incidence and milk protein content at



around day 60 post-calving ( $60 \pm 5$  days). However lactation number was not included during the analysis of first lactation heifers. The metabolic weight, which is related to energy expenditure (basal metabolic rate) was calculated by raising the body calving weight to the power 0.75 (Terpstra, 2001). Calving ease had two variables; 0 for cows with normal calving and all other categories were classified as 1. In an event where a cow was served more than once after first calving, only the last recorded BCS at service was included in the analysis. The trait genetic line had a variable 0 for select and 1 for control. Based on the feeding system, low forage diet were assigned variable 0 and high forage diet 1.

Age at first calving and calving weight have been shown to affect subsequent animal performance, for instance on milk production capacity and longevity (Wathes et al., 2007). It is also a recommendation for Holstein-Friesian dairy heifers that their age at first calving should be around 24 months at a liveweight of approximately 550 kg (Kennedy et al., 2010). Hence it was important to include age at first calving and calving weight in the analysis. In dairy cows, health problems, lowered reproductive performance, reduced milk yield, feed intake and milk production have all been associated with altered body condition in the animals studied (Ferguson et al., 1994). For example, cows with excessive body condition loss end up having reduced fertility (Mulligan et al., 2007). As such, BCS at service was added during the analysis. Calving ease in cows is a risk factor for impaired subsequent reproductive performance (Price and Wiltbank, 1978) and on a financial basis dystocia can impact fertility by 34% of production costs (Mee, 2007). Hence calving ease was also selected for subsequent analysis. The amount of milk protein at around day 60 of lactation was included because metabolic disorders can be reflected in altered biochemical composition of milk content (Cejna and Chladek, 2005). In particular, milk protein is linked with energy status of dairy cows, with its proportion in milk decreasing during periods of energy deficit (Tena-Martinez et al., 2009). Dobson et al., (2008) further stated that mastitic cows have lowered conception rates compared to healthy cows, hence mastitis was included in the subsequent analysis.

#### **2.2.2.1.2 Traits evaluated for cows culled due to udder problems**

Traits considered to contribute to an increased risk of culling cows due to “lactation disease” problems were: peak milk flow rate, duration of milking, days to peak milk yield, lactation number and lactation diseases. This variable “lactation diseases” had two variables. Variable 1 if the cow had history of being diagnosed with any of the following: teat blockage and contusion, hard quarters, teat injury, retained placenta, ketosis and milk fever in the lactation culled. Variable 0 was otherwise.

The variable peak milk flow rate was used as a proxy for teat canal diameter, which is one of the udder characteristics which are not expected to change on a short time scale (Chagunda et al., 2006). In addition to this, duration of milking is an index reflecting the negative effect of machine milking function on teat integrity (Chagunda et al., 2006). It is also known that udder problems tend to increase with parity, and one of the risk factors for culling cows due to udder problems include teat problems (Milian-Suazo et al., 1988). Further some of the infectious (teat injury and metritis) and non-infectious (retained placenta, ketosis and milk fever) diseases increase the risk of mastitis (Chagunda et al., 2006), possibly via the resulting physiological stress. Retained placenta alone can in some cases result in a decreased average daily milk yield (Ganah et al., 2008). The number of days to peak lactation was included in the analysis to represent the stress experienced by cows during early lactation. Knop and Cernescu, (2009) indicated that during early lactation, cows mobilise body reserves to sustain milk production. Hence a hypothesis was made that the time it takes for a cow to reach her peak lactation can be related to the degree of stress being experienced by the cow.

#### **2.2.2.1.3 Traits evaluated for cows culled due to accident**

Locomotion score and lactation number were the traits that were in used to assess the increased risk of culling cows due to accident in the final model. Locomotion score had two variables; 1 for cows with a score of 1 and 2 (ie. normal and mild lameness), and 2 for all cows with a score of greater or equal to 3 (ie. severe lameness) in the

lactation the cow was culled. Control cows were assigned a variable based on observations in the whole lactation. The scores were classified so that variable 1 should represent animals which were normal and mildly lame, while variable 2 animals were easily recognised to be lame. There is some inconsistency to this scoring system, for example sometimes cows with big udders are given a score of 2 rather than 1 because there is no uniformity in gait.

The locomotion scoring system assesses animal gait to detect lameness in the animals (Sprecher et al., 1997). On this basis, locomotion score reflected difficulties in movement which affected cow activity. The severity of an injury that a cow can suffer depends on her situation or the kind of activity at the time of accident; hence it was selected to be one of the independent variables. Due to differences observed in the number of cows culled per lactation, lactation number was also included in the statistical model.

### 2.2.3 Statistical Analysis

In order to determine the predisposing factors for culling dairy cows due to identified major reasons, a backward stepwise multiple regression model was applied. The analysis was conducted using logistic regression applying both CATMOD and logistic procedure of SAS version 9.2 (SAS, 2008). Through CATMOD procedure, the predicted probabilities of culling cows were generated while odds ratio estimates were obtained through logistic procedure. Logistic regression was used because the dependent variable, the fate of being culled, was a discrete variable with only two outcomes. The logit was the response function which is defined as  $\text{Log}\left(\frac{PY=0}{PY=1}\right)$  (SAS, 2008). The response variable Y (the fate of either being culled or surviving) had the value 1 for cows that were culled, and value 0 otherwise. The following multiple regression model (Moskowitz and Wright, 1985) was applied:

$$Y_i = \beta_0 + \sum_{j=1}^K \beta_j X_{ij} + \varepsilon_i \quad (i = 1 \text{ and } 0)$$

Where:  $Y_i$  =  $i^{\text{th}}$  observation of the dependent variable.

$K$	=	number of independent variables.
$x_{ij}$	=	$i^{\text{th}}$ observation of the $j^{\text{th}}$ independent variable.
$\beta_0, \beta_1, \dots, \beta_K$	=	unknown constants.
$\epsilon_i$	=	$i^{\text{th}}$ disturbance term.

To identify the model that fitted the evaluated traits, likelihood ratio goodness-of-fit test in the CATMOD procedure output was used. Calculations of predicted probability were based on the formula shown below (SAS, 2008).

$$P = \frac{e^{E(\text{logit}(p))}}{1 + e^{E(\text{logit}(p))}}$$

Where: $P$	=	Predicted probability.
$E(\text{logit}(p))$	=	Predicted value of the logit.

The model automatically excluded all the records with missing values in one of the traits. This resulted in only cows with complete records being included in the analysis.

## 2.3 Results

### 2.3.1 Descriptive Statistics

Cows assessed in this study for an increased risk of being culled due to various reasons had an average age at first calving of 26.2 (SD = 2.4) months. The minimum age at first calving was 22.2 months and the maximum was 37.8 months.

### 2.3.2 Reasons for Culling

There were 519 cows which had their first calving between September 2003 and August 2010. A total of 175 of these cows were culled for various reasons. The classified reasons for which the cows were culled are presented in Table 2-1.

Table 2-1: Reasons for culling in Holstein-Friesian dairy cows.

Reason for culling	Total culled	Total % rate	1 <sup>st</sup> lactation culls	1 <sup>st</sup> lactation % culling rate	1 <sup>st</sup> lactation culls as a percentage of all lactation culls
Infertility	48	9.2	23	2.5	47.9
Udder problems	47	9.1	13	2.1	27.6
Accidents	32	6.2	11	1.0	34.3
Unknown or Others	20	3.9	5	0.4	25.0
Foot or leg problems	22	4.2	2	0.4	9.0
Died	6	1.2	2	10.8	33.3
Culled	175	33.7	56	4.4	32
All cows	519				

The first three reasons for culling in the herd were infertility (9.2%), udder problems (9.1%) and accident (6.2%). Another common reason for culling cows was foot or leg problems (4.2%), which was common especially in multiparous cows as compared to first lactation heifers. Hence infertility, udder problems and accident were identified as the major reasons for culling in the studied Holstein-Friesian herd. Narrowing to major reasons, within infertility 87.5% were culled because the cows were repeat breeders, 10.4% had reproductive disorders and 2.1% were not seen in heat. Within udder problems, 53% were culled due to mastitis, 17% poor udder conformation, 15% lost quarter, 11% high SCC and 5% damaged udder. Combining mastitis and high SCC they accounted for 64% of all cows culled due to udder problems. Based on housing system, more cows (18) were culled due to accident from the group of cows housed throughout the year than the group of cows which had access to fields in summer (14).

Table 2-2 details the number of cows culled based on lactation number in the Holstein-Friesian dairy herd for the first three lactations a cow was supposed to stay in the production systems project.

Table 2-2: Number of cows culled per lactation in the Holstein-Friesian dairy herd.

Lactation	Reasons for culling						Total number of cows culled	Percentage culled of total number of culls	Percentage culled of total number of cows
	Infertility	Udder problems	Accident	Foot/leg problems	Died	Others			
One	23	13	11	2	2	5	56	32	10.8
Two	10	8	8	5	1	7	34	19.4	7.5
Three	7	9	9	7	3	3	38	21.7	7.3
All cows Culling rate								175	519
									<b>33.7</b>

In general, the percentage of culled cows per lactation decreased with an increase in lactation number. Additionally, 10.8% of all cows which had their first calving within the study period did not progress to their second lactation. The predominant reason for culling cows in their first lactation was infertility (23). Looking at the proportion of cows culled per lactation, it appears to suggest that overall the risk of culling cows decreased with increasing parity number.

Culling rates based on the four production systems showed a higher culling rate in the LFS system, both overall (11.2%) and in first lactation heifers (3.5%). The second and third highest culling rate by production system was for LFC (9.2%) and HFS (7.1%) systems. The lowest culling rate was 6.2% in HFC system. Thus more high genetic merit cows were culled, in particular cows fed the low forage diet regardless of parity number.

### 2.3.3 Predisposing factors for cows culled for each of the three main reasons

#### 2.3.3.1 Cows culled due to infertility

Descriptive statistics for cows studied for increased risk of being culled due to fertility reasons are presented in Table 2-3.

Table 2-3: Descriptive statistics of cows evaluated for increased risk of culling cows due to infertility.

Trait	Descriptive Statistics	Culled	Control
	Mean (Standard Deviation)	26.1 (3.0)	25.9 (2.5)
Age at first calving (months)	Min	22.2	22.7
	Max	36.6	33.1
	Mean (Standard Deviation)	119.5 (12.0)	119.6 (12.0)
Metabolic calving weight (kg)	Min	95.7	94.1
	Max	143.5	146.2
	Mean (Standard Deviation)	1.2 (0.3)	1.3 (0.2)
Milk protein content around day 60	Min	0.6	0.9
	Max	1.9	1.9
	Mean (Standard Deviation)	1.2 (0.3)	1.3 (0.2)

The average age at first calving for cows which were culled due to infertility was slightly higher (26.1 months, SD = 3.0) than their matched cohorts (25.9 months, SD = 2.5). The median (2.25) and third quartile (2.5) BCS at service were also higher for cows which were culled due to infertility than control (2 and 2.25, respectively), but not first quartile (culled = 1.75 and control = 2).

The final regression model for cows culled due to fertility reasons had four traits (Likelihood ratio = 0.05). The traits were: calving ease, BCS at service, mastitis and 60d milk protein content. The BCS at service and 60d milk protein content showed a

significant impact ( $P < 0.05$ ) on culling in dairy cows due to infertility. The regression estimate for BCS at service was positive (1.67) and for 60d milk protein content was negative (-2.43), presented in Table 2-4.

Table 2-4: Likelihood estimates for the increased risk of culling cows due to infertility.

Parameter	Estimate	P-Value
Intercept	-0.54	0.76
Calving ease	-0.62	0.30
Body condition score at service	1.67	0.03
Mastitis	0.98	0.20
Milk protein content at around day 60 (kg)	-2.43	0.02

Therefore cows with a high BCS at service and low 60d milk protein content were at an increased risk of being culled for infertility. The maximum predicted probability for this model was 0.91 given calving ease = 1, BCS at service = 3, mastitis = 0 and 60d milk protein content = 0.6. The odds ratios for calving ease, BCS at service, mastitis and 60d milk protein are in Table 2-5.

Table 2-5: Odds Ratio of the traits included in the model used to predict the increased risk of culling cows due to infertility.

Trait	Estimate	95% Wald Confidence limit	
		Lower	Upper
Calving ease	0.5	0.18	1.7
Body condition score at service	5.3	1.15	24.3
Mastitis	2.7	0.55	11.9
Milk protein content at day 60 (kg)	0.1	0.01	0.7

The odds ratio for BCS at service was 5.3. This means that cows with a higher than average BCS at service were 5.3 more times likely to be culled. Cows with low milk protein at around 60 days in milk were 10 times less likely (a risk of 9.1%) to be culled.



For the first lactation heifers culled due to fertility problems, the regression model had four traits (Likelihood ratio = 0.05). The traits were: age at first calving, metabolic calving weight, BCS at service, genetic line and feeding system. However of all the tested traits, BCS at service was the only trait that had significant influence ( $P < 0.05$ , regression estimate = 2.65 and predicted probability = 0.86) on the likelihood of cows being culled in their first lactation due to fertility reasons, Table 2-6.

Table 2-6: Likelihood estimates for the increased risk of culling first lactation heifers due to infertility.

Parameter	Estimate	P-Value
Intercept	-5.78	0.26
Age at first calving (months)	0.06	0.64
Metabolic calving weight (kg)	-0.02	0.63
Body condition score at service	2.65	0.02
Genetic line	-0.20	0.83
Feeding system	0.95	0.22

The maximum predicted probability estimate was based on a sample with age at first calving = 23, metabolic calving weight = 94.1, BCS at service = 2, genetic line = 0 and feeding system = 0 (this would be a cow in the LFS group which had the highest culling rate by production system). Table 2-7 shows the odds ratios of the traits in a fitted model of first lactation heifers culled due to fertility reasons.

Table 2-7: Odds Ratio of the traits included in the model used to predict the increased risk of culling first lactation heifers due to infertility.

Trait	Estimate	95% Wald Confidence limit	
		Lower	Upper
Age at first calving (months)	1.07	0.8	1.4
Metabolic calving weight (kg)	0.98	0.9	1.1
Body condition score at service	14.14	1.4	143.1
Genetic line	0.82	0.1	5.1
Feeding system	2.58	0.57	11.7

First lactation heifers with a high BCS at service were more vulnerable (Odds Ratio 14.1) to the risk of being culled for fertility reasons. This highlights fertility as being a predominant reason of culling cows in their first lactation in the present study

### 2.3.3.2 Cows culled due to udder problems

The descriptive statistics of some of the traits for cows culled due to udder problems are in Table 2-8.

Table 2-8: Descriptive statistics of some of the traits evaluated for an increased risk of culling cows due to udder problems.

Trait	Descriptive Statistics	Culled	Control
	Mean (Standard Deviation)	37 (16.9)	62 (35.0)
Days to peak lactation (days)	Min	7	18
	Max	89	185
	Mean (Standard Deviation)	4 (1.5)	4.2 (1.1)
Peak milk flow rate (Lmin <sup>-1</sup> )	Min	1.5	2.2
	Max	7.5	7
	Mean (Standard Deviation)	9.8 (3.6)	9.7 (2.7)
Duration (min)	Min	4	6
	Max	18	19

The mean interval from calving to peak lactation for cows which were culled for udder problems was lower (37 days, SD = 16.9) than that of matched cows (62 days, SD = 35.0). The maximum number of days to peak lactation for culled cows was also lower (89 days) than that of their control counterparts days (185).

Among the six traits in a fitted model (Likelihood ratio = 0.1), days to peak milk yield had a significant influence ( $P < 0.01$ , regression estimate = -0.05 and probability = 0.89) on an increased risk of culling cows due to udder problems, shown in Table 2-9.

Table 2-9: Likelihood estimates for the increased risk of culling cows due to udder problems.

Parameter	Estimate	P-Value
Intercept	2.44	0.151
Lactation number	-0.18	0.462
Days to peak lactation (days)	-0.05	0.003
Peak milk flow rate (Lmin <sup>-1</sup> )	0.02	0.921
Duration of milking (min)	0.01	0.916
Lactation diseases	-0.21	0.975

The probability predicted was when lactation number = 1, days to peak milk yield = 7, peak milk flow rate = 2.4, duration of milking = 8 and lactation diseases = 0. The odds ratios are in Table 2-10.

Table 2-10: Odds Ratio of the traits included in the model used to predict an increased risk of culling cows due to udder problems.

Trait	Estimate	95% Wald Confidence limit	
		Lower	Upper
Lactation number	0.84	0.5	1.4
Days to peak lactation (days)	0.96	0.9	1.0
Peak milk flow rate (Lmin <sup>-1</sup> )	1.03	0.6	1.7
Duration of milking (min)	1.01	0.8	1.2
Lactation diseases	0.81	0.2	3.8

Cows with short or long intervals to peak lactation had equal odds (approximately 1) of being culled for udder problems. Thus although the number of days to peak

lactation was significant, the risk of culling cows with udder problems due to a short interval from calving to peak lactation was the same as that of the cohorts.

### 2.3.3.3 Cows culled due to accident

Summary statistics of the data set analysed for a raised risk of culling cows due to accident are presented in Table 2-11.

Table 2-11: Descriptive statistics of cows evaluated for an increased risk of culling cows due to accident.

Trait	Descriptive Statistics	Culled	Control
	Mean (Standard Deviation)	22.3 (2.9)	22.7 (3.5)
Growth rate (kgmonth <sup>-1</sup> )	Min	17.0	18.7
	Max	29.3	31.2
	Median	2	2.5
Locomotion score (rank)	First quartile	1.5	2
	Third quartile	3	3.75

The mean growth rates show that culled (22.3 kgmonth<sup>-1</sup> SD = 2.9) and control (22.7 kgmonth<sup>-1</sup> SD = 3.5) cows were not different. For locomotion score, control cows had higher values for median, first and third quartiles than cows culled due to accident.

The fitted model for cows assessed for an increased risk of being culled due to accident had two independent variables; locomotion score and lactation number. None of these had a significant influence ( $P > 0.05$ ) on the likelihood of culling cows due to accident.

## 2.4 Discussion

In the present study, infertility (9.2%), udder problems (9.1%) and accident (6.2%) were identified to be the major reasons for culling in a Holstein-Friesian dairy herd. Similar results have been reported previously (Bascom and Young, 1998; Cozler et al., 2009; Esslemont and Kossaibati, 1997 ; Milian-Suazo et al., 1988), with fertility still standing as the most important reason for culling in dairy cows. From the literature, reported culling rates (Milian-Suazo et al., 1988) showed reproduction (4.8%), udder problems (4.0%) and low production (3.8%) to be the major reasons for culling cows. With so many factors now involved in making a decision to cull a cow (Bascom and Young, 1998), it was suggested that cows are rarely culled due to low milk yield alone (Esslemont and Kossaibati, 1997 ). The fact that infertility is consistently the major reason for culling in both previous and current studies could be due to a number of factors including feeding management, and also because reproductive performance is highly influenced by the genetic value of the cows. Studies have shown that high producing cows tend to be more susceptible to body energy imbalances as measured by excessive body condition loss (Heuer et al., 2000; Wathes et al., 2007). Udder problems came second in the current study because their classification combined mastitis, lost quarters, high SCC, damaged udder and poor udder conformation, whereas in other studies these reasons were reported separately. These reasons were combined in the present study due to the small total number of cows culled in each individual category. In the literature, mastitis is reported as an independent reason of culling, for example by Esslemont and Kossaibati, (1997). In another study, Bascom and Young, (1998) ranked mastitis second as a reason for culling, with poor udder conformation seventh and SCC eleventh. This is likely to explain the differences between the previous studies and this present study, as in the present study udder problems was the second major reason for culling. Further despite having a low reported culling rate for accident (0.3%) (Milian-Suazo et al., 1988), it is always listed (Bascom and Young, 1998; Cozler et al., 2009) amongst the reasons of culling in dairy cows. This would indicate that it is still a significant reason for culling that should not be overlooked. However not all studies indicate accident as a reason for culling, with Bascom and Young (1998) indicating death

(not accident) while Cozler et al., (2009) indicated accident (not death) as a reason for culling. In the present study, these two reasons were not combined because cows which were culled due to accident referred to cows that suffered a severe injury and later ended up being culled. In other words, these were cows culled due to injury, excluding injuries associated to foot or a leg which were recorded separately as foot and leg problems.

Within the major reasons for culling identified in the current study, it was shown that the proportion of primiparous cull cows was high (47.9% of the total cows culled for fertility reasons were 1<sup>st</sup> lactation heifers). For cows culled due to udder problems, the proportion of primiparous culled cows was 27.7%, and for accident 34.4%. A study by Cozler et al., (2009) indicated that most of the heifers were culled before the end of the trial because of reproductive problems after their first calving. In a different study (Frelich et al., 2010) it was indicated that culling for fertility reasons was common in parity one and two. Estimation for culling rates in first lactation heifers can be as high as 33% (Wathes et al., 2007). Thus fertility problems appear to be significant in first lactation heifers. These results illustrate the importance of fertility management not only in 1<sup>st</sup> lactation heifers, but also the importance of fertility problems generally in the dairy industry.

In the current study, having infertility as an outstanding reason for culling could be attributed to the fact that the reproductive performance of cows depends on factors which are both not influenced and highly influenced by management. For example a factor such as the genetic value of cows does not depend on management, but calving body condition is highly dependent on management. Data used in the current study included cows which were selected based on genetic merit, and further subgrouped by feeding systems. In addition to this, the cows were under an intensive management system because they were on a long term genetic and feeding systems project (Bell and Roberts, 2006; Chagunda et al., 2009). The impact of negative energy balance on fertility is significant in high producing cows, because such cows tend to lose more body condition (Knop and Cernescu, 2009; Mulligan et al., 2007; Wathes et al., 2007). Hence despite being on highly controlled feeding management

(ad-lib TMR), their risk of being culled was also potentially affected by their genetic value. Genetic merit could be a contributing factor to the high culling rate observed in LFS production system. The greater impact in first lactation heifers could be attributed to the fact that they were still growing (as compared to cows in second or later lactations), hence this would represent an increased challenge for first parity cows. Furthermore negative energy balance can elevate SCC and mastitis incidence (Banos et al., 2006), which could have possibly helped increase the risk of being culled due to udder problems in the present study. Most of the cows culled due to udder problems were mainly culled because of high SCC and mastitis. On the other hand, the numerical difference observed in the number of cows culled due to accident between group of cows housed throughout the year and cows which had access to field in summer could be due to housing condition, although there was no evidence to support this assumption. Housing is mostly associated with an increased lameness incidence (Rushen and de Passille, 2006), as well as increased levels of handling which may predispose to an accident (Waiblinger et al., 2004). However further work on investigating cows culled due to accident is necessary.

The reasons for culling in the present study have shown that most of the dairy cows are culled due to fertility reasons and udder problems. In addition, involuntary culling is also influenced in rare cases by accident. This knowledge is very important for our understanding of the current culling pattern and management of cows in dairy herds (Hadley et al., 2006). Current culling pattern is vital because it helps to make informed decisions for culling on the farm. On the other hand, management in terms of feeding is important because it has an influence on how dairy cows meet their production demands, with the aim of reducing physiological stress from body energy deficit especially in high producing cows.

Cows with a high BCS at service and low 60 day milk protein had an increased likelihood of being culled due to infertility, regardless of their parity. The maximum predicted estimated probability for that event to occur was 0.91. Culling was estimated to occur approximately 5.3 times more often for cows with higher than average BCS. From the literature, it has been shown that excessive body condition in

dairy cows is not desirable as it is associated with increased health problems (Ferguson et al., 1994). Cows with higher than average BCS score ( $\geq 3.5$ , 5-point scale) are prone to metabolic disorders (Roche et al., 2009), for instance development of fatty liver syndrome (Heuer et al., 1999). Fatty liver syndrome is a metabolic disorder which is associated with other metabolic diseases (eg. ketosis, retained placenta and milk fever) or udder diseases specifically mastitis in early lactation (Roche et al., 2009; Shearer and Van Horn, 1992). Regarding ketosis, it has been shown that cows developing ketosis tend to have a higher BCS than healthy cows, and a history of ketosis before service decreases the likelihood of conception to that service (Gillund et al., 2001). Retained placenta on the other hand poses a great challenge to reproductive performance of cows because it reduces conception rate (Ganah et al., 2008). Mastitis has been reported to affect reproduction by increasing the number of days open for cows diagnosed with clinical mastitis after first service (Loeffler et al., 1999). Incidences of milk fever are rare in first- and second-lactation animals, but gradually increase from third to sixth parities (Shearer and Van Horn, 1992). In advanced stages, milk fever may even lead to the death of an animal. Overall there is an indication that metabolic disorders contribute to an increased susceptibility of cows to health challenges which compromises cow fertility. Regarding first lactation heifers, this study has shown that only higher than average BCS at service had a significant influence on the likelihood (probability = 0.86) of culling primiparous cows for fertility reasons. High BCS at service after first calving in this study was demonstrated to be a disadvantage to primiparous cows, because it made them 14.1 times more likely to be culled for fertility reasons. Therefore there is an assumption that there are additional challenges for first lactation heifers compared to at least second parity cows. One previous study (Mee, 2007) indicated that body condition outside the BCS range of 2.75-3.5 (5-point scale) in first lactation heifers increased the risk of calving difficulties. From other studies, the effects of dystocia include impaired reproduction (Price and Wiltbank, 1978) and a greater risk of being culled, with greater impact in parity one than subsequent parities (Lopez et al., 2007). Early lactation is a critical time for dairy cows because it is when cows are required to establish a good reproductive performance (Minor et al., 1998). In addition during



early lactation, intakes of the diet may be unable to support the increased demand for nutrients resulting in excessive mobilisation of body reserves (Ross et al., 2008).

One of the potential reasons for cows having a higher than average body condition score at service in the current study was that the animals were on highly controlled production systems, which reduced the chance of having significant numbers of thin cows by the time of service. In addition the body condition of cows was assessed on a weekly basis, hence most of the cows were likely to have calved in good body condition. However due to differences in rates of body condition loss in respect to individual production demand and the ability to withstand the physiological stress during early lactation, such issues with body condition must have affected the level of body condition loss or gain during their early lactation period. Animals culled because of not being seen in heat, were repeat breeders and had reproductive disorders were classified as being culled for infertility. Excluding those culled due to a reproductive disorder, these other reasons contributed approximately 90% of the total cows culled due to infertility. Generally body condition score increases with time in lactation (Roche et al., 2009). Hence there was an increased probability of serving cows with high body condition score in repeat breeders, and those not seen in heat. In addition, the BCS at service which was used in the current study was the last recorded BCS, not the first if the cow did not conceive after first service for other unrecorded reasons. This might explain why more cows with higher than average BCS at service were culled for infertility reasons.

Cows were also at risk of being culled due to infertility if they had a low milk protein at around day 60 in lactation. The risk of culling cows with low milk protein content at around 60 days was 9.1%. A study by Tena-Martinez et al., (2009) reported that cows producing milk with a lower protein percentage than average were more likely to be culled by the end of their lactation. Literature shows that there is a relationship between milk composition and energy status of a cow (Vries and Veerkamp, 2000). Furthermore metabolic disorders can affect the biochemical characteristics of milk, for example a fat to protein ratio higher than 1.5 may be indicative of subclinical ketosis and energy deficit (Cejna and Chladek, 2005). A different report indicated

that in times of energy deficit, the protein proportion in milk tends to decrease (Tena-Martinez et al., 2009). In circumstances where a cow experiences severe negative energy balance, it has been reported that reproduction performance in later lactations can be compromised (Collard et al., 2000; Heuer et al., 2000) through adverse effects on follicular development, ovarian activity and elevated mastitis incidence (Banos et al., 2006; Leroy et al., 2008). Contributing factors leading to negative effects on follicular development and ovarian activities have been found due to alterations in GH and a decline in IGF-1 (Maurice and Lonergan, 2003; Wathes et al., 2007) amongst other effects. In some cases severe negative energy balance can also result in an animal not conceiving at all and hence being culled (Knop and Cernescu, 2009).

In contrast to previous studies, the current study used the amount of protein at around day 60 in lactation rather than the proportion of protein in milk. Ratio of milk fat to milk protein was not used, because of the strong correlation with milk protein which was used. Despite these difference in methodology, similar results were obtained. The correlation between 60d milk fat and 60d milk protein was positive, while the correlation between 60d milk protein and ratio of 60d fat to 60d milk protein was negative. Thus the decrease in milk protein was associated with a decrease in milk fat as well, but proportionally milk protein was decreasing at a lower rate than milk fat which was reflected by the negative correlation between protein and ratio of fat to protein. On around day 60, these early lactation cows would still be susceptible to energy deficit. In early lactation nutrient demand is high, and high producing cows are vulnerable to entering a state of negative energy balance (Knop and Cernescu, 2009). Bewley and Schutz, (2008) indicated that although the primary body reserve mobilised is fat during a time of energy deficit, cows may also tap into reserves of protein or minerals. Hence it was suggested that cows with low amounts of milk protein at around day 60 in lactation were at an increased risk of being culled for fertility reasons.

The stress on the udder before peak lactation was associated with cows that had a short duration to peak milk yield. Cows with a reduced interval to peak milk yield in

the present study were at an increased risk of being culled for udder problems. The odds ratio for days to peak lactation showed that cows with a short interval to peak lactation were no more likely to be culled for udder problems compared with controls. In a different study (Milian-Suazo et al., 1989), it was predicted that cows would be culled for udder problems if they had lower previous-lactation milk per day, higher current milk per day, clinical mastitis and teat problems. According to Banos et al., (2006), negative energy balance elevates SCC and mastitis incidence. In the current study 64% of the cows classified as being culled for udder problems were culled due to high SCC and mastitis. The hypothesis for the significance of number of days to peak lactation on the risk of culling cows due to udder problems is that it reflected an increased stress on the udder, which might have an influence on the incidences of mastitis and high SCC.

The results of this study on factors that predisposed cows to an increased risk of being culled due to infertility and udder problems have demonstrated the importance of feeding and consistent monitoring of body condition of dairy cows. Feeding management ensures that animals maintain good body condition at each stage in lactation, while routine body condition scoring ensures that animals are not over conditioned. Hence both activities can help to mitigate the challenge of high producing cows entering into a state of severe negative energy balance. According to Roche et al., (2009), all cows can optimally produce in respect to their genetic and production systems without compromise to reproduction, welfare and health within the range of acceptable body condition score profiles.

Locomotion score and lactation number were not significant on increasing the likelihood of culling cows due to accident. However traits describing the temperament of cows were not included in the present study. Including temperament parameters in the analyses could have possibly given different results. Previous studies by Waiblinger et al., (2004) showed that defensive reactions of animals due to fear, pain or startling increase the risk of injuries to these animals.

## **2.5 Conclusion**

This study has demonstrated that infertility, udder problems and accident are the main reasons for culling dairy cows in the Langhill herd. The study has also shown that a higher BCS than average at service, low milk protein content around day 60 in lactation and short duration to peak lactation had an influence on the vulnerability of cows to being culled. In particular, higher than average BCS at service and low milk protein content at around day 60 in lactation increased the risk of culling cows due to fertility reasons, potentially due to their associations with metabolic disorders and body energy deficit respectively. Reduced interval to peak lactation was related to an increased susceptibility of cows being culled due to udder problems.

### **Chapter 3**

#### **Effect of Temperament on Days to First Recorded Oestrus in Dairy Cows**

### **3.1 Introduction**

Temperament is amongst the behavioural characteristics that are observable in dairy cows. In farm animals, temperament is often described as an individual trait influencing an animal's behavioural response to handling (Gibbons et al., 2009). However, some authors have described it as how an individual reacts to novel or challenging situations (Denis et al., 2000). Handling of animals has been shown to have an influence on growth rate and pregnancy rate in gilts, and milk production in cows (Boissy and Bouissou, 1988). Rough handling at service on the other hand has been associated with lowered conception rates because of resultant stress (Waiblinger et al., 2004). There is also a reported association between the flight distance kept by an animal to milk yield, though contrasting results have been reported (Munksgaard et al., 2001). In addition, there is an indication of a relationship between the flight distances kept to a person by an animal, with an animal's level of fear determined by flight scores (Munksgaard et al., 2001). The genetic estimates between temperament traits and breeding traits show positive correlation, for example that less fearful heifers are more productive based on higher reproduction indices and calving performance (Phocas et al., 2006). Temperament is also being included in computation of selection indexes amongst traits such as milk yield, calving ease, fertility, mastitis resistance and milking rate that breeders take into account (Kristensen, 1995). However in order for temperament tests to be feasible for use on commercial farms (Gibbons et al., 2009), it is necessary to be able to test the animal in its home environment without removing the animal from its social group. Gibbons et al., (2009) stated that this principle has been demonstrated by evaluating human approach and avoidance tests in the home environment, for example at the feed-face and whilst lying. Boissy and Bouissou (1988) also demonstrated this principle by scoring flight distances even on lying animals, by approaching the cows quietly in a standardised way.

### **3.1.1 General Objective**

The overall objective of this study was to determine the effect of temperament on oestrus in Holstein-Friesian dairy cows.

#### **3.1.1.1 Specific Objectives**

The specific objectives were:

1. To determine the difference in temperament between primiparous and multiparous Holstein-Friesian dairy cows.
2. To find out if temperament changed over time in Holstein-Friesian dairy cows.
3. To determine the effect of temperament on days to first recorded oestrus in Holstein-Friesian dairy cows after calving.

### **3.2 Materials and Methods**

#### **3.2.1 Animals**

The study used the Scottish Agricultural College (SAC) Langhill herd of Holstein-Friesian cows. It was conducted at the SAC Dairy Research Centre located in Dumfries, Scotland, United Kingdom. The animals were on a long-term genetic and feeding systems project (Bell and Roberts, 2006). Under genetic line, the cows were classified as either select line or control line; based on feeding systems, the animals were either fed on a high or low-forage diet as a total mixed ration (Chagunda et al., 2009). The project therefore had four groups of cows based on the genetic and feeding systems. The groups were: high forage control, high forage select, low forage control and low forage select. No changes were made to how the cows were managed at the farm. The routine handling experienced by the cows was predominantly handling in the milking parlour and during body condition assessment. The cows were milked three times a day, and body condition scoring was done on a weekly basis. Prior to first calving, all the heifers were managed together. The heifers were

first served at between 14 and 15 months of age, in order to calve down at an age of around 24 months. After calving, the cows were served from at least 42 days post-calving.

### **3.2.2 Experimental Design and Data**

Seventy Holstein-Friesian dairy cows were monitored in this study. Thirty five of these were primiparous cows, and the other half multiparous cows. To be included in the study, a cow had to be at least 30 days precalving at the time of the first test. The pairing of the cows was based only on parity; primiparous and multiparous. Four additional cows were available, to be replacements in case of dropouts from the sample groups. Each animal was subjected to a modified Approach Test (AT) described by Gibbons et al., (2009). A detailed protocol is in Appendix A. The aim of the test was to create a situation where the cow was given space to express her response to human approach (Gibbons et al., 2009). According to Gibbons et al., (2009) the test is consistent in respect to time, and thus the only type of test capable of indicating some of the ideal factors of temperament. The test was carried out at three distinct times (A, B and C) on each animal during the experimental period. The test was carried out on day  $30 \pm 3$  before calving (A), and on  $30 \pm 3$  and  $60 \pm 3$  days after calving (B and C, respectively). The period of the experiment was from May 2010 to December 2010. The temperament traits of interest were: flight response score, nervousness, interest, shyness, boldness, fearfulness and docility. The behaviour traits were observed by one assessor. Data used to calculate the number of days to first recorded oestrus after calving was sourced from SAC Langhill database approximately a month after completing the AT test. The hypothesis was that an increased time to first oestrus indicated possible fertility challenges, as the number of ovulatory cycles preceding service positively correlates to conception (Knop and Cernescu, (2009).



### **3.2.3 Statistical Analysis**

A total of 188 records were available from 62 of the 70 tested animals. Some of the animals did not complete the experiment because they were either transferred out of the production systems or culled, mostly animals in multiparous group. All of the statistical analysis were carried out using SAS version 9.2 (SAS, 2008). In the analysis, flight response score represented the temperament of cows. This trait was selected because the protocol used to estimate nervousness, interest, shyness, boldness, fearfulness and docility had a high potential of confounding more factors than that used for flight response. In practice it is easier to estimate flight response score as compared to nervousness, interest, shyness, boldness, fearfulness and docility.

#### **3.2.3.1 Temperament Traits**

To find out if there was a statistical difference in flight response scores between the primiparous and multiparous cows, the Friedman test was used to calculate Cochran-Mantel-Haenszel statistics (based on rank scores) by applying the Frequency (FREQ) procedure. Factors taken into account in the analysis were genetic merit, feeding system and experimental area (Cubicles and Field). Experimental area unfortunately could not be controlled during data collection. For example heifers had the first test at time A in the field, and B and C either in the field or cubicles depending on the system allocated. Cows in high forage groups had almost all of the tests at time A, B and C done in the field. Further analysis was conducted to find out if the temperament score in individual cows did change from time A to C. The null hypothesis tested was that there was no significant difference between the flight response scores at period of test A, B and C. In the analysis for location, the null hypothesis used Wilcoxon signed rank test by applying the UNIVARIATE procedure. Since flight response score was selected to represent the temperament traits, the flight response score was correlated to nervous, interest, shy, bold, fear and docile. Spearman correlation coefficients were calculated by applying the Correlation (CORR) procedure to identify the nature of the relationship between the flight

response score and nervous, interest, shy, bold, fear or docile. The individual data set for temperament traits and the differences between the periods was not normally distributed hence the use of nonparametric analysis procedures.

### 3.2.3.2 Temperament and oestrus

After studying the behaviour of the temperament traits, the effect of flight response score on days to first recorded oestrus was assessed. In order to determine whether the temperament of the cows affected the number of days to first recorded oestrus after calving, analysis of variance (ANOVA) applying the Generalised Linear Models (GLM) procedure was conducted. Data for days to first recorded oestrus after calving were normally distributed. The independent variables in the model were genetic merit, feeding system, cow group and flight response score at time A, B and C.

## 3.3 Results

### 3.3.1 Descriptive Statistics

Overall, interest had the highest mean score 107.3 (SD = 19.2) than the mean for nervous 73.3 (SD = 45.2), shy 53.3 (SD = 46.3), bold 64.6 (SD = 46.9), fear 56.4 (SD = 47.8) and docile 59.9 (SD = 45.4), presented in Table 3-1.

Table 3 -1: Descriptive statistics of some of the traits studied in the Langhill herd.

Description (In millimetres unless indicated)	Mean	SD	Minimum	Maximum
Nervous	73.3	45.2	0	125
Interest	107.3	19.2	10	125
Shy	53.3	46.3	0	125
Bold	64.6	46.9	0	125
Fear	56.4	47.8	0	125
Docile	59.9	45.4	0	125
Time to first recorded oestrus (days)	55.5	17.9	23	101
Age at first calving (months)	25.4	1.9	22.5	31.7

In addition, interest was the only temperament trait which was present in all cows. The minimum score for interest was 10 while the rest of the qualitative temperament traits had both extremes observed. On average, the number of days to first oestrus after calving was 55.5 days (SD = 17.9). The minimum number of days to first recorded oestrus after calving was 23 days, and maximum was 101 days. The animals had an average age at first calving of 25 (SD = 1.9) months. Thus the animals calved for the first time at an age close to the target.

### 3.3.1.1 Flight Response Score

The frequency distribution of flight response score of the primiparous cows and multiparous cows on a 9-point ordinal scale is shown in Figure 3-1.

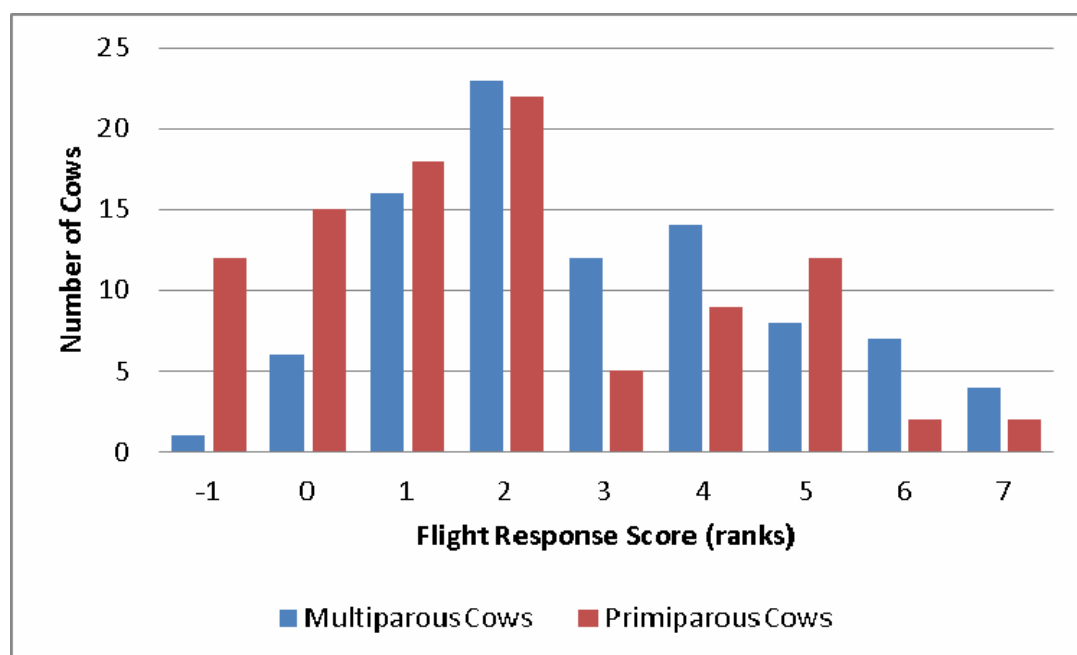


Figure 3-1: Distribution of animals on 9-point flight response score ordinal scale.

*Ranks: -1, Flight Distance (FD) > 3m; 0, FD ≤ 3m but > 2m; 1, FD ≤ 2m but > 1m; 2, FD ≤ 1m but > 0m; 3, FD = 0m; 4, FD = extends arm to touch; 5, FD = touches the cow's head; 6, FD = touches cow's body/rump and 7 Cow moves towards an experimenter.*

The distribution of scores showed that primiparous cows tended to move away from the experimenter at a longer flight distance than multiparous cows. This is shown by

the highest frequencies of flight response score in ranks -1, 0 and 1. On the other hand, multiparous cows responded to an experimenter at a shorter flight distance than primiparous cows. The multiparous cows had the highest frequencies in all ranks of greater or equal to 2 (excluding the rank of 5). However both groups had the highest frequency at a flight distance when an experimenter was less or equal to a metre but not getting in contact; which was scored a rank of 2. One cow from the multiparous group had a rank of -1, giving the lowest observed rank in that group. The lowest frequency number for primiparous cows was 2, observed in ranks 6 and 7.

The average ranks of the flight response score for the two groups of cows based on period of test are shown in Figure 3-2.

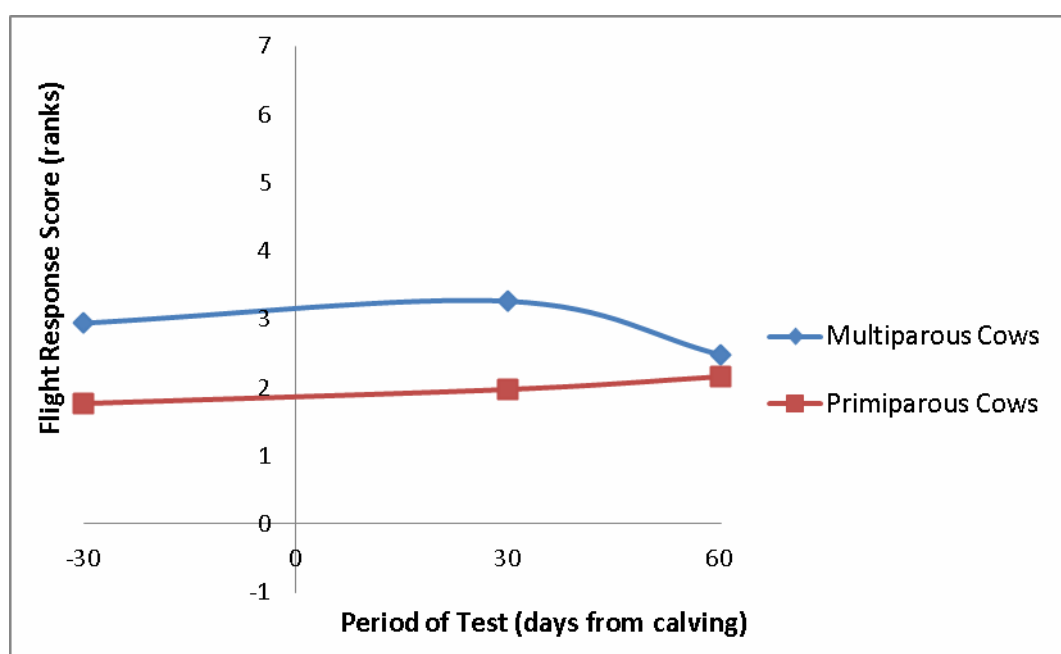


Figure 3-2: Flight response score averages at day -30, +30 and +60 from calving.

The Figure 3-2 clearly shows that multiparous cows overall moved away from the experimenter at a shorter flight distance than primiparous cows, because multiparous cows had the highest average rank flight response score in all three periods. The overall difference was significant ( $P < 0.05$ ). The average flight response score (ranks) for multiparous cows at time A was 3.06, and 1.59 for primiparous cows. During time B both increased in average rank score, multiparous cows reaching 3.1

and primiparous cows 2.03. However during time C, the graph shows that multiparous cows increased flight response distance because the average rank dropped to 2.48, whilst flight distance decreased in primiparous cows since the average rank score increased to 2.16. In general, the first lactation heifers had a steady decrease in flight distance from time A to C.

The comparisons of flight response scores for multiparous and primiparous cows in all three periods of test are shown in Figure 3-3.

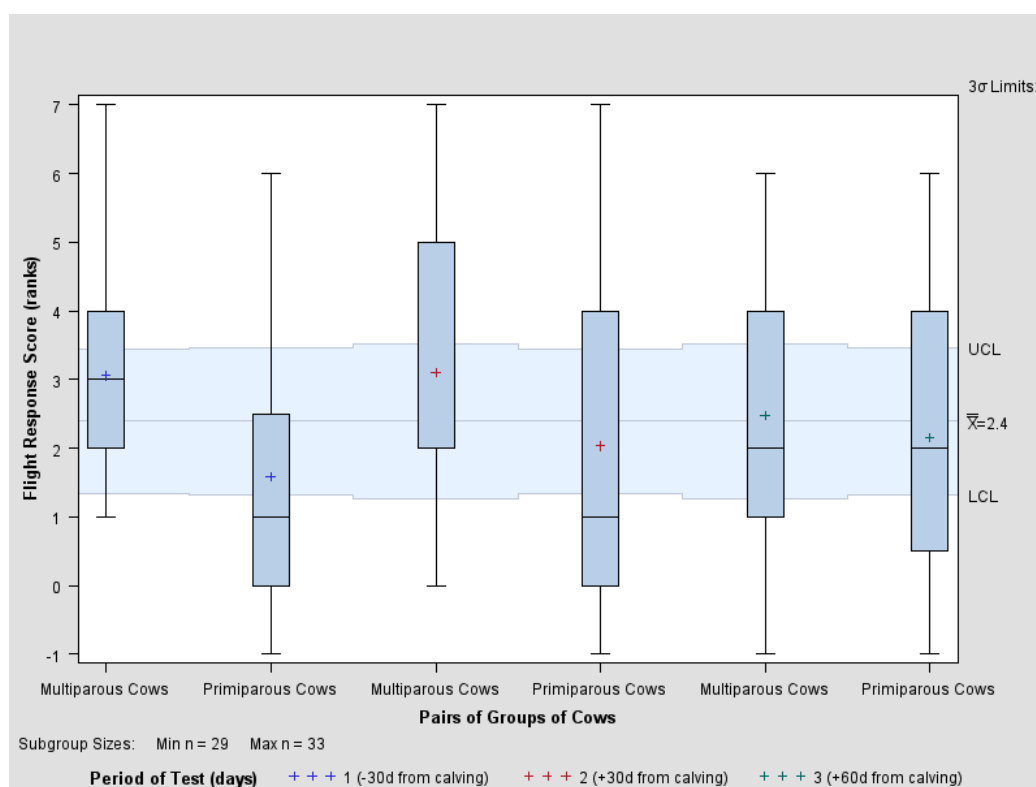


Figure 3-3: Comparisons of flight response scores between primiparous and multiparous cows at time A (1), B (2) and C (3).

*Flight response score rank: -1, flight distance (FD) > 3m; 0, FD ≤ 3m but > 2m; 1, FD ≤ 2m but > 1m; 2, FD ≤ 1m but > 0m; 3, FD = 0m; 4, FD = extends arm to touch; 5, FD = touches the cow's head; 6, FD = touches cow's body/rump and 7 Cow moves towards an experimenter. UCL= upper control limit and LCL = lower control limit.*

The median flight response score for multiparous cows was 3 at time A, and 2 at time B and C. Primiparous cows had median flight response score of 1 at time A and B,

and 2 at time C. In general, Figure 3-3 shows that the two groups were significantly different ( $P < 0.05$ ) at time A and B, compared to time C. The 25% quartile 1 (Q1) and 75% quartile 3 (Q3) for multiparous cows was: 2 and 4, 2 and 5, and 1 and 4 for time A, B and C, respectively. Primiparous cows had similar Q3 for time B (4) and C (4), but different to that of time A (2.5). Similarly the Q1 for primiparous cows was 0 for both time A and B, while time C had Q1 of 0.5. From the graph it is also shown that multiparous cows at time B had the same value for both the median and Q1.

### 3.3.2 Temperament Traits

Cochran-Mantel-Haenszel statistics (based on rank scores) showed that there was a significant difference ( $P < 0.05$ ) in flight response score between the primiparous cows and multiparous cows. Therefore it implied that primiparous cows were different from multiparous cows in the way they responded to approaching humans during the Avoidance Test. However the null hypothesis that there was no significant difference between the three periods A, B and C in flight response score was supported ( $P > 0.05$ ). Statistically cow temperament as measured by flight response score did not change from time A to C.

Table 3-2 presents the Spearman's correlation coefficients ( $r_s$ ) for flight response score and six qualitative temperament traits: nervousness, interest, shyness, boldness, fearfulness or docility.

Table 3-2: Spearman's rank correlation coefficients of temperament traits.

	Flight Response Score	P-value
Nervous	-0.71	< 0.001
Interest	-0.07	0.329
Shy	-0.76	< 0.001
Bold	0.79	< 0.001
Fear	-0.80	< 0.001
Docile	0.83	< 0.001

The Table 3-2 shows that flight response score was significantly ( $P < 0.001$ ) correlated to nervous, shy, bold, fear and docile. The results were not far from expectation, because these qualitative traits are inherently dependent on the flight response of the cow as judged by the assessor. An increase in flight response score was associated with an increase in the levels of boldness ( $r_s = 0.79$ ) and docility ( $r_s = 0.83$ ) and a decrease in the level of nervousness ( $r_s = -0.71$ ), shyness ( $r_s = -0.76$ ) and fearfulness ( $r_s = -0.8$ ) of cows.

### 3.3.3 Temperament and Oestrus

Assessment of the effect of temperament on days to first recorded oestrus showed that genetic merit, feeding system, cow group and flight response score at time A, B and C did not significantly ( $P > 0.05$ ) affect onset of oestrus after calving, shown in Table 3-3.

Table 3-3: Probability values of the independent variables in the model used to determine the effect of temperament on onset of oestrus after calving.

Variable	F-value	P-value
Cow group	0.87	0.36
Genetic merit	0.72	0.41
Feed system	0.36	0.55
Flight response score at time A	0.27	0.97
Flight response score at time B	0.44	0.89
Flight response score at time C	0.51	0.82

From the Table 3-3 it is shown that at both the 1% and 5% confidence level, the onset of first oestrus after calving was not affected by the temperament of cows.

### 3.4 Discussion

The overall objective of this study was to relate cow temperament to number of days to first recorded oestrus in dairy cows. The results showed that the number of days to first recorded oestrus after calving was not significantly affected by the flight response score. In other words, the occurrence of first recorded oestrus did not depend on the temperament of cows. In a different study, it was indicated that reduced stress during service might enhance the non-return rate and rough handling at service resulted in lowered conception rate (Waiblinger et al., 2004). According to Robert and Mathew, (2000) factors related to stress stimulate secretion of epinephrine and norepinephrine as part of the fight-or-flight reaction (Robert and Mathew, 2000). However Inest et al., (2002) demonstrated the influence of catecholamines on the ovaries in frogs, and found that norepinephrine can be one of the factors responsible for the metabolic processes that characterise cytoplasmically immature oocytes. Therefore it was suggested that temperament might have negative effects on the physiological activities of the reproductive system, especially after service. Possibly rough handling or the animal's response to rough handling at service can stimulates secretion of catecholamines, which are capable of interfering with other biological processes occurring between service and conception. With that assumption, it was suggested that the use of a different fertility trait (for example days open) might be another approach to quantify the effect of cow temperament on fertility traits because oestrus does not reflect the success (or otherwise) of a service event. After calving, there is less likelihood of stressful handling to cows except in the case of complications that might happen at or after calving. In the present study, interval from calving to conception was not included in the analysis because the number of cows which were in-calf was not sufficient.

The flight response scores did not significantly change from time A to C, supporting the null hypothesis. According to Gibbons et al., (2009) a trait must be consistent in its behaviour over time and across situations in order to be classified as a temperament trait. Therefore by definition of temperament, the results from the current study are consistent with this description. Gibbons et al., (2009) also reported



that flight response scores measured by human approach test were consistent in three subtest repeats. The Gibbons et al., (2009) study repeated the test over an 11 day period separated by a minimum of 2 days, while in the current study the period was approximately 90 days and the tests were separated by 60 days (time A and B) and 30 days (time B and C). However there is no defined time period that can be used as a benchmark for repeatability in such behavioural testing, hence the period of this experiment (approximately 90 days) might have contributed to the variation in results. In addition only one experimenter was involved in estimating the flight response scores, which may have affected the variations in scores. Rousing et al., (2005) reported that calves during on-farm testing of behaviour responses were able to distinguish between a stockperson and unfamiliar test person in their test response. Hence there would be possibly more variations in flight response scores if more than one experimenter were involved.

However there was a significant difference ( $P < 0.05$ ) in flight response scores between the primiparous cows and multiparous cows in the current study. The differences in frequency distribution showed that most of the primiparous cows responded at a longer distance than multiparous cows to an approaching human during experimental tests by moving at least two steps away. A clear difference was shown by differences in averaged ranks of flight response score, subgrouped by periods of test and comparison plots at time A and B. The steady increase in average flight response score for primiparous cows from time A, B and C suggested that overall the flight distance at which primiparous cows responded to an experimenter was reducing with time. A sudden increase in flight distance was observed for multiparous cows at time B to C. No clear reason could be ascertained for this, but it is speculated that this change could be due to maternal related factors. Reports in the literature suggest that the change in primiparous cows over time may be related to prolonged or previous handling in animals which has an influence on man-animal relationships (Boissy and Bouissou, 1988) or restless behaviour of an animal (Waiblinger et al., 2004). Additionally first parity cows tend to be more nervous, but then get calmer in later lactations (Fuerst, 2006). Through extrapolation, it is possible to predict that the group mean flight response scores would converge after calving

and more repeated handling of primiparous animals. Thus outside time C, it would be expected that the observed difference between primiparous and multiparous cows would be reduced.

In the current study, the source of differences in flight response score between the primiparous and multiparous cows was highly likely to have been influenced by a history of previous handling of the animals. However the location of where the test was conducted (in a field or cubicles) could also be a possible confounding factor. The difference in handling between the two groups was that primiparous cows were not often handled before calving, compared to multiparous cows which were handled more often especially during milking and body condition scoring. All lactating cows were milked three times a day and body condition was assessed once a week by palpation. Hence multiparous cows were handled more often than first lactation heifers prior to their first calving. Comparisons based on a time scale show that multiparous cows were at least a year ahead of primiparous cows in terms of experiencing extra handling (for example during milking and body condition scoring). All primiparous cows at time A were still pregnant heifers, and at time C they were around day 60 in lactation. One possible explanation for the steady decrease in overall flight response distance was due to the influence of routine handling during milking and body condition scoring after calving. Before calving, the heifers were predominantly handled as a group not individuals. Apart from this, in heifers the nature of human contact and familiarity affects the subsequent response to humans (Breuer et al., 2003). Technically the test employed in the current study was positive, and it was conducted by one experimenter.

With interest to the welfare of animals, it is important to know if there are differences in cow temperament at different growth or production stages. This knowledge is helpful because it can assist in improving handling techniques, so that there is reduced stress on cows during handling (Waiblinger et al., 2004).

Further results indicated that flight response score was significantly ( $P < 0.01$ ) correlated to the qualitative temperament traits. Flight response score represented

temperament of the cows in the present study, because it is easier to estimate flight response score than other qualitative temperament traits. Comparatively the protocol used to estimate flight response score was less likely to confound other factors than the protocol used to estimate the qualitative temperament traits. The significant relationship between flight response score and the individual five qualitative temperament traits was strong (coefficients greater than 0.7). Increase in flight response score in the present study was associated with a decrease in nervousness, shyness and fearfulness and an increase in boldness and docility of dairy cows. Munksgaard et al., (2001) indicated that the flight distance kept by an animal to a human is often used as a measure of fear. Fear of humans in animals can be an accumulative source of stress (Rousing et al., 2005). Factors related to stress (for instance anticipation of danger) stimulate secretion of hormones (epinephrine and norepinephrine) as part of fight-or-flight reaction (Robert and Mathew, 2000). Therefore one possible source of the difference in how nervousness, shyness and fearfulness relate to flight response score (as compared to boldness and docility) would be because these traits stimulate differential secretion of hormones by the adrenal gland. Nervousness, shyness and fearfulness in dairy cows can be related to situations which may cause dairy cows to anticipate danger, or to be fearful if handled roughly.

Results on the relationship between flight response score and qualitative temperament traits have shown that the distance at which a cow responds to humans (avoidance distance) can be associated with nervousness, shyness, fearfulness, boldness and docility of dairy cows. This was likely to be the case in the present study, since most of these behavioural traits are inter-related. However it was suggested by Munksgaard et al., (2001) that the relationship between distance of movement and level of fear in animals is complex, in the sense that it may depend on situation rather than a primary sign of fear. However the results from the current study have shown that to some extent the flight distance can be related to levels of nervousness, shyness, fearfulness, boldness and docility in dairy cows. Thus animals can easily be identified to be fearful, docile or nervous based on the avoidance distance and so should be handled with extra care. Fearful animals on the farm are

likely to be negatively affected by recurring contact with humans (Rousing et al., 2005) if they are not properly handled. In addition, human avoidance and the Approach Test have been supported as assessors of the quality of human-animal relationship in dairy cows (Rousing et al., 2005).

### **3.5 Conclusion**

This study has demonstrated that cow temperament did not affect days to onset of oestrus after calving. However there was a difference in flight distance between primiparous and multiparous cows around calving. Further results suggested that avoidance distance is potentially a suitable means of estimating levels of behaviour related traits (nervousness, shyness, fearfulness, boldness and docility) in dairy cows.

## **Chapter 4**

### **General Discussion**

#### **4.1 Vulnerability in dairy cows**

The present work has shown that a higher than average BCS at service and low milk protein content at around day 60 in lactation expose cows to an increased risk of being culled due to fertility reasons. Short duration to peak lactation is correlated with an increased susceptibility of culling cows due to udder problems. The challenge with a higher than average BCS at service is that it increases the risk of developing metabolic disorders, some of which then negatively affect conception rate (Gillund et al., 2001; Roche et al., 2009). However there is a great challenge for first lactation heifers because heifers enter into first lactation before they are mature enough to withstand the physiological stress of early lactation. Although reports indicate a low proportion of milk protein to be an indicator of energy deficit (Tena-Martinez et al., 2009; Vries and Veerkamp, 2000), in the present study it has been shown that a low amount of protein content in the milk at around day 60 in lactation may also relate to negative energy balance and so reduced fertility. In addition, the effects of negative energy imbalance and the physiological stress that cows experience during early lactation may increase levels of mastitis and high SCC (Banos et al., 2006), which were identified as the major reasons for culling cows due to udder problems.

These results suggest that consistent feeding and monitoring of body condition in dairy cows is of paramount importance in reducing how vulnerable cows are to culling for infertility. Feeding management is vital because it helps to ensure that animals calve down in good body condition, while accompanied by routine body condition scoring to ensure that animals are not over-conditioned. Whilst emphasis is placed on calving body condition, these results suggest that BCS at service is also of great importance. However maintenance of optimal body condition at each stage of lactation can be of great help (Wildman et al., 1982). In addition, first parity cows appear to need extra care as the current study has shown that these cows were highly vulnerable to an increased risk of being culled due to fertility reasons.

Through monitoring of primiparous and multiparous cows over a period of approximately 90 days around calving, it has been demonstrated that the temperament of primiparous cows as measured by flight response score is different from that of multiparous around calving. The difference observed was proposed to be a consequence of lactating multiparous cows experiencing extra handling during routine human contact such as milking and body condition scoring. However after calving, first lactation heifers appear to get more used to humans as indicated by a steady decrease in avoidance distance as demonstrated in the current study and as reported by Fuerst, (2006). Despite the differences in temperament between primiparous and multiparous cows, temperament did not significantly change which is consistent with literature (Gibbons et al., 2009). Thus temperament traits do not change over a short time period. On the other hand, the present study has shown that the flight distances of cows can be related to how nervous, fearful, shy, bold and docile the cows are, indicating the possibility of quantifying qualitative temperament traits based on avoidance distance. In addition the current study has shown that the temperament of cows did not delay the onset of first oestrus after calving. However some authors suggest that temperament may be more influential at service (Waiblinger et al., 2004).

Understanding the behaviour of temperament traits in dairy cows is helpful in improving the welfare of cows. If cows can be identified with abnormal temperament in dairy herds, then such cows may need to be handled accordingly to reduce stress, which in addition may enhance their reproductive performance (Waiblinger et al., 2004) and human-animal relationship (Rousing et al., 2005). Furthermore in order to address the question of whether cow temperament has an influence on fertility, the use of different fertility traits (for example days open) has been proposed. To fully address the effects that cow temperament might have on reproductive performance, detailed research on how physiological changes arise due to different levels of temperament traits may be required, in particular at service.

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## **Appendix**

### **A. Approach Test Protocol<sup>1</sup>**

The test was carried out by one experimenter wearing a red fleece with blue overall. The criteria for starting this test was that the focal cow had to stand idle in the passageway of the housing area or field, with sufficient space to move away from the experimenter and have no more than 2 cows standing within 1m. When these criteria were fulfilled, the experimenter approached the cow from a distance of greater than 3m in a standardised way. The experimenter approached the focal cow using strides of approximately 1m and after every step the experimenter remained motionless for 10 seconds to allow the cow to respond. The experimenter approached diagonally from the front towards the cow's neck, avoiding eye contact with the cow, looking towards the feet of the cow and keeping arms and hands close to the body. Avoidance was recorded using a flight response score which is defined as the distance at which the cow responds by taking two or more steps in the opposite direction from the approaching experimenter. In some cases cows shuffled their feet or take a half step or even a full step in the opposite direction but the experimenter

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<sup>1</sup> As detailed by Gibbons et al., (2009) with a few modifications

continued approaching the cow. Likewise, the experiment continued when cows turn their heads in the opposite direction. The distance was measured by eye at an approximate distance of 1m in the field and using cubicle width (approx. 1m) as a guide in cow house. The flight response score was measured on a 9-point ordinal scale (Table A-1).

Table A-1: The flight response score used to score the cow's flight response to the AT test.

Score	Behavioural response
-1	Cow moves away when experimenter is $> 3\text{m}$
0	Cow moves away when experimenter is $\leq 3\text{m}$ but $> 2\text{m}$ away
1	Cow moves away when experimenter is $\leq 2\text{m}$ but $> 1\text{m}$ away
2	Cow moves away when experimenter is $\leq 1\text{m}$ but $> 0\text{m}$ away
3	Cow moves away when experimenter is $0\text{m}$ away
4	Cow moves away as experimenter extends arm to touch
5	Cow moves away as experimenter touches the cow's head
6	Cow does not move away as experimenter touches the cow's head/body/rump
7	Cow moves towards an experimenter

Source: Gibbons et al., (2009).

On completion of the test, a qualitative assessment was made of the cow's response (see Table A-2). The experimenter marked an individual visual analogue scale (VAS) for six qualitative terms, according to a subjective judgement of whether a cow scored low or high for each term.



Table A -2: Qualitative terms and descriptions used in the AT test.

Term	Description
Nervous	An animal that is quite restless/wary/uneasy as the experimenter approaches. May avoid experimenter or turn head in opposite direction. The animal may quiver/flinch when a hand is placed on her.
Interest	An inquisitive or playful animal that is very alert of the experimenter approaching and/or other events happening around her.
Shy	A timid animal that is easily frightened. An animal that is distrustful, suspicious, reluctant, wary, lacks confidence. The animal may draw back or avoid experimenter.
Bold	A bold animal does not hesitate, is confident and may approach experimenter. May be very inquisitive and try to sniff/lick/rub experimenter.
Fear	An animal that is afraid, anxious, apprehensive and uneasy. A fearful animal will avoid experimenter.
Docile	A docile animal is easily approached and handled. Appears comfortable and/or calm as experimenter approaches.

Source: Gibbons et al., (2009).

The VAS consisted of 125mm horizontal line with two vertical lines marking the extreme points of the scale (0 mm: term absent, 125 mm: term present throughout the test). Scores for each term were measured as the distance in millimetres from the 0-point.